GENETIC PORTRAIT OF 12 X-STR LOCI IN BRAZIL IMMIGRANT POPULATION LIVING IN LISBON (PRELIMINARY RESULTS)

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Introduction

Typing X-chromosome markers (ChrX) is important to complement the analysis of Y and autosomal (AS) short tandem repeats (STR). The main field of the ChrX is kinship investigations, especially investigations of mother/son, father/daughter and complex kinship tests. When the alleged father is not present, the ChrX investigation is more accurate because the mean exclusion chance of X-STRs in these cases is higher. ChrX markers testing also prove helpful for kinship tests in which only remote relatives are available for testing, particularly from the need to rejoin families in the context of war, mass disasters and world-wide migration. Since male family members are more likely to be affected by the consequences of socio-demographic unrest than females, and it may therefore be more difficult to obtain samples from them, ChrX marker testing could be a very useful tool.

Immigrant populations contributes to increase social, cultural, religious, linguistic, anthropological and genetic heterogeneity in a population. According to the portuguese database PORDATA [1], in 2016, the number of immigrants from Brazil who have fixed residence in Portugal was 79,569. From those, up to 40.084 live in Lisboa and near cities. Consequently, we are therefore faced with a population reality completely different from the one at the early ninety's of the last century. Before forensic application, it´s important to study population data and to construct reference databases to document the genetic variation of specific STR among worldwide populations. Population data is vital to quantify the evidentiary value of a match in forensic evaluation.

The aims of this study were to calculate allele frequencies and create a population database for X-STR markers for Brazil natives, living in Lisboa for future use in forensic genetics practice.

Material and Methods

Population genetic data of 12 X-STRs, (DXS10074, DXS10079, DXS10101, DXS10103, DXS10134, DXS10135, DXS10146, DXS10148, DXS7132, DXS7423, DXS8378 and HPRTB) were analysed in 50 males of an immigrant population from Brazil living in Lisbon. Since the studied X-STRs genetic markers are distributed on four linkage groups (LG) (Table 1), that can provide independent genotype information, we studied those groups as haploytes.

Extracted samples [2] were amplified using the Investigator Argus X-12 kit (Qiagen) with half-volume reactions (12.5 µL) following the manufacturer's guidelines.

Samples were run on a 3130xlGenetic Analyzer (Thermofisher Scientific) following the manufacturer's protocol. Fragments were sized using the internal size standard BTO-550 and genotyped using the allelic ladder provided with the kit (Qiagen). All off-ladder alleles were determined by size and re-injected for confirmation. Raw data was analyzed using GeneMapper ID-X 1.4 (Thermofisher Scientific).

The software ARLEQUIN 3.5 [3] was used to calculate allele frequencies of the 12 X-STR loci. Polymorphism information content (PIC) power of discrimination in females (PD_F), power of discrimination in males (PD_M), mean exclusion chance in trios involving daughters (MEC_T) and mean exclusion chance in father/daughter duos (MEC_D) calculations, for each X-STR, were performed using http://www.chrx-str.org/ [4].

Results

Several out off-ladder alleles were observed at DXS10146 system (alleles 22, 26.2 and 37.2); and at DXS10148 system (alleles 14, 17, 28 and 32.1).

Forensic statistical parameters for each X-STR considering 50 male genetic profiles are shown in Table 1. PIC ranged from 0.60832 (DXS7423) to 0.92790 (DXS10135). The power of discrimination for females ranged from 0.83194 (DXS7423) to 0.99128 (DXS10135), while the PD for males ranged from 0.66240 (DXS7423) to 0.93200 (DXS10135).

The combined power of discrimination (PDs) for the four triplets of X-STR loci in females and males as well as the combined mean exclusion chance in trio cases involving daughters and in father/daughter duo cases are shown in Table 2.

When examining the population data as haplotypes, linkage group I shown to be the most informative, reviling 48 unique haplotypes and the less polymorphic group was linkage group III reviling 34 unique haplotypes (Table 3).

	DSX10103	DXS8378	DXS7132	DXS10134	DXS10074	DXS10101	DXS10135	DXS7423	DXS10146	DXS10079	HPRTB	DXS10148	
PIC	0,66410	0,65570	0,62257	0,83686	0,86917	0,89736	0,92790	0,60832	0,91204	0,79953	0,74321	0,88417	PIC
PD_F	0,87248	0,86143	0,84461	0,96408	0,97416	0,98350	0,99128	0,83194	0,98765	0,94507	0,91659	0,97988	PD_F
PD_M	0,70400	0,71040	0,66720	0,85040	0,88080	0,90480	0,93200	0,66240	0,91760	0,82320	0,77680	0,89280	PD_M
MEC_T	0,66410	0,65570	0,62257	0,83686	0,86917	0,89736	0,92790	0,60832	0,91204	0,79953	0,74321	0,88417	MEC_T
MEC_D	0,52053	0,51106	0,47597	0,73408	0,77818	0,82114	0,86963	0,46157	0,84453	0,68237	0,89280	0,80155	MEC_D

Table 1. Forensic statistical parameters for each X-STR in a sample of 50 males of the Brazil immigrant population living in Lisboa.

	Linkage Group I	Linkage Group II	Linkage Group III	Linkage Group IV
PD_F	0.99998	0.9998	0.9998	0.99992
PD_M	0.998	0.993	0.994	0.996
MEC_T	0.997	0.990	0.991	0.994
MEC_D	0.99	0.96	0.99	0.98

Table 2. Population genetic data of relevant forensic efficiency p arameters for four triplets of X-STR markers in Brazil immigrant population living in Lisboa.

Linkage group	Linkage Group I	Linkage Group II	Linkage Group III	Linkage Group IV
Number of different haplotypes	49	45	41	45
Number of unique haplotypes	48	41	34	41

Table 3. Compiled data on haplotypes of the four linkage groups of Cabo Verde immigrant population living in Lisboa.

Conclusions

The obtained results for forensic statistical parameters such as polymorphism information content, power of discrimination and mean exclusion chance, based on single allele frequencies, reveal that this multiplex system is highly informative and can represent an important tool for genetic identification purposes in the immigrant population of Brazil.

In the present study DXS10135 is the most polymorphic marker and DXS7423 is the marker that presents less diversity. The forensic efficiency parameters for four closely linked groups were all higher than 0.96, with linkage group I being the most polymorphic and linkage group II the less informative.

The obtained forensic statistical parameters suggest that this four linkage group are highly informative and represent an important tool for genetic identification purposes in general researches.

References

[1] - Pordata, Fevereiro de 2018, available at: www.pordata.pt.

[2] - P. Walsh, D. Metzger, R. Higuchi. Chelex 100 as a médium for simple extraction of DNA for PCR-based typing from forensic material. Biotechniques. 1991; 10(4):506-13.

[3] - L. Excoffier, G. Laval, S. Schneider. Arlequin (version 3.0): An integrated software package for population genetics data analysis. Evolutionary Bioinformatics Online. 2005 (1):47-50.

[4] - Szibor R, Hering S, Edelmann J (2006) A new Web site compiling forensic chromosome X research is now online. Int J Legal Med 120(4):252–254. doi:10.1007/s00414-005-0029-y

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